242-Pos
Comparative Study of the Effect of UV- VS. Gamma Radiation on Human Hair
Ervin Palma[1], David Gomez[1], Eugene Galicia[2], **Yuri V. Griko**[3].
[1]San Jose State University, San Jose, CA, USA, [2]Eloret Co., Sunnyvale, CA, USA, [3]NASA Ames Research Center, Mountain View, CA, USA.
The effect of UV- and ^{60}Co gamma radiations on the structural and chemical integrity of human hair has been studied to determine the feasibility of using human hair as a non-invasive biomarker of radiation exposure to ionized and non-ionized radiation. The goal was to identify the most sensitive molecular parameter associated with radiation-induced damage and to evaluate effects of dose range from 0.5Gy to 10Gy. Steady state tryptophane fluorescence and chemical analytical methods were utilized to quantitatively evaluate molecular integrity of Trp fluorophor and SH-groups in hair proteins to asses the radiation induced damage.
The study found that human hair fibers were progressively damaged by exposure to both UV- and ionized gamma radiation up to 10Gy. Damage to the hair was evidenced by a decrease in the fluorescence intensity due to depletion of the amino acid tryptophan as well as significant reduction in a number of free SH- groups.
The results show that hair-fibrils exposed to gamma rays of higher quantum energy than UV, undergo much smaller extent of changes in Trp fluorescence and SH-group modification, then when exposed to equal energy of UV-irradiation. The changes appear faster under the effect of UV than by gamma-irradiation and their intensity and character depend on the dose. The stable Trp fluorophore is extremely sensitive to UV-B and UV-C radiation in contrast to the ionized gamma radiation, which causes damage from the reaction of free radicals and direct deposition of energy.
We conclude that fluorescence intensity of the hair is a sensitive parameter for the quantitative evaluation of the radiation exposure.

243-Pos
Direct Observation of Ostwald Ripening in Free-Interface Diffusion Based Protein Crystallization
Aaron M. Streets[1], Stephen R. Quake[1,2].
[1]Stanford University, Stanford, CA, USA, [2]Howard Hughes Medical Institute, Chevy Chase, MD, USA.
In the late stages of first order phase transitions, interfacial energy minimization drives competitive growth between precipitates of the new phase. This dissolution of small precipitates to feed the growth of larger clusters is commonly referred to as Ostwald ripening. Here, Ostwald ripening is directly observed during the late stages of Lysozyme crystallization in a microfluidic free-interface diffusion reactor. In order to measure the wide range of crystal sizes necessary to quantitatively characterize the ripening phenomenon, we developed a novel dynamic light scattering apparatus, with an incorporated microscope, based on the microfluidic crystallization device. The crystal size distribution mean, observed by scattering, grows as $t \perp 1/3$ in agreement with the analytic predictions of Lifshitz Wagner and Slyozov (LSW). In the case where a large crystal emerges outside of the scattering volume, local crystal size distributions deviate from power law growth in an expected fashion. A simulation based on LSW theory is implemented to model the crystal size distribution evolution. The observed growth trajectories show quantitative agreement with simulated kinetics.

244-Pos
Conformational Flexibility of Aggregation-Prone Peptides Studied By PET-FCS
Sören Doose[1], Marc Löllmann[2], Michael Schwering[3], Markus Sauer[1].
[1]Julius-Maximilians University Würzburg, Würzburg, Germany, [2]Bielefeld University, Bielefeld, Germany, [3]Ruprecht-Karls University Heidelberg, Heidelberg, Germany.
Glycine-serine polypeptides in aqueous solution behave as unstructured polymers that exhibit end-to-end contact rates dependent on contour length and in accordance with polymer theory. Aggregation-prone peptides like polyglutamine (poly-Q, associated with various neurodegenerative diseases) of sufficient length were shown to form more collapsed states in aqueous solution. Conformational dynamics mediated by intrachain, interchain, and chain-solvent interactions can reveal mechanistic aspects for the aggregation process that is thought to be responsible for the onset of disease. We used contact-induced fluorescence quenching due to photoinduced electron transfer (PET) between terminally attached oxazine fluorophores and tryptophan for studying peptide loop closure rates on ns-µs time scales. Conformational dynamics as function of temperature, solvent viscosity, and osmolyte concentrations were analyzed using fluorescence correlation spectroscopy (PET-FCS). Significantly smaller end-to-end contact rates for poly-Q and related peptides as compared to poly-GS peptides with a viscosity-dependence hinting at internal friction were found.

Membrane Protein Structure I

245-Pos
X-Ray Structure Determination of Isocytochrome C_2 from the Photosynthetic Bacterium *Rhodobacter sphaeroides*
Peace C. Esonwune[1,2], Herbert L. Axelrod[3].
[1]San Francisco State University, San Francisco, CA, USA, [2]Stanford University- Stanford Synchrotron Radiation Lightsource, Palo Alto, CA, USA, [3]Stanford University, Palo Alto, CA, USA.
In cytochrome c_2-deficient strains of the photosynthetic bacterium, *Rhodobacter sphaeroides*, a related c-type cytochome, isocytochome c_2 replaces cytochrome c_2 as the secondary electron donor to the photo-oxidized bacteriochlorophyll dimer in the reaction center. We will describe the x-ray structure determination of isocytochrome c_2 at 2.2 Å from *Rb sphaeroides.*Analysis of the x-ray diffraction data indicated the crystals belong to space group P3(2) with unit cell dimensions a=b=56.0 Å c=90.5 Å and are merohederally twinned. The structure of isocytochrome $c_2$from *Rb. sphaeroides* shows significant structural similarity to the cytochrome c_2 from a different photosynthetic bacterium, *Blastochloris viridis*.

246-Pos
Structure Dynamics and Allosteric Regulation of the E. Coli High-Affinity Methionine Transporter Metni
Eric Johnson, Chris Vercollone, Allen T. Lee, Douglas C. Rees.
Howard Hughes Medical Institute and Division of Chemistry and Chemical Engineering, California Institute of Technology, Pasadena, CA, USA.
The high-affinity uptake of methionine by *Escherichia coli* is mediated by MetNI, a member of the methionine uptake transporter family of ATP-binding cassette (ABC) transporters. We previously reported the crystal structure of MetNI at 3.7 Å resolution and a brief analysis of the methionine mediated trans-inhibition of the transporters' ATPase activity[1]. Here, we report two new crystal structures of the MetNI transporter, solved at 2.8 and 4.0 Å resolution. While both structures of MetNI reveal the transporter adopting an inward-facing conformation, significant changes are observed in the conformation of the MetI transmembrane, MetN nucleotide binding, and MetN-C2 carboxyl-terminal regulatory domains. The conformational changes can be described primarily as rigid-body movements which result in a partial closing of the MetN nucleotide binding domains, and a simultaneous rotational rearrangement of the MetN-C2 regulatory domain. The kinetic properties of trans-inhibition have also been characterized by an analysis of ligand binding on the ATPase activity of wild type and specific MetN-C2 regulatory domain mutants.
1. Kadaba N, Kaiser J, Johnson E, Lee A, and Rees D C. The high affinity *E. coli* methionine ABC transporter: structure and allosteric regulation. Science. 2008 Jul 11;321(5886):250-3.

247-Pos
Betp - X-Ray Structure and Function of An Osmosensor and Transporter
Reinhard Kraemer[1], Susanne Ressl[2], Vera Ott[1], Sascha Nicklisch[1], Heinz-Juergen Steinhoff[3], Lucy Forrest[2], Christine Ziegler[2].
[1]University of Koeln, Koeln, Germany, [2]Max-Planck-Institute of Biophysics, Frankfurt, Germany, [3]University of Osnabrück, Osnabrueck, Germany.
The Na+ coupled betaine uptake system BetP of Corynebacterium glutamicum belongs to the BCCT family of transporters and comprises both a catalytic function (betaine/Na+ cotransport) and a sensory/regulatory function responding to osmotic stress. Its 2D structure was recently solved by electron and its 3D structure by X-ray crystallography. Within a trimeric structure, each BetP monomer harbours both an N- and a C-terminal domain involved in stimulus sensing and intramolecular signal transduction. Factors contributing to the sensory and regulatory function of BetP are (i) the two terminal domains, (ii) K+ ions as an osmostress related stimulus, and (iii) interaction with the surrounding membrane.
We used several techniques to analyze the contribution of the terminal sensory domains to BetP function. By scanning mutagenesis we identified the significance of single amino acids and parts of the C-terminal domain of BetP. EPR spectroscopy was applied to determine the mobility or the C-terminal domain under different functional conditions and to measure intra- and intermolecular distances in BetP. The 3D structure of BetP finally revealed a putative crosstalk between the three monomers of BetP via their C-terminal domains. In addition, the observed asymmetry of the 2D crystal demonstrates the presence of different conformational states of the three individual monomers.
On the basis of these studies we suggest a novel functional model of intersubunit crosstalk between the three individual monomers as well as the terminal domains of BetP during its catalytic and its sensory function.